***yeaRp* Experiment Protocol**

Growth Medium: Luria Broth (LB)

The test samples were first grown in LB overnight at 37^{o}C. They were then washed with 0.85% NaCl solution. Washed samples were then resuspend into different concentrations of medium in a 96-well deep well plate and were further grown at 37^{o}C until the bacteria reached mid-log phase. The fluorescence output was then measured using an EnVision multilabel reader.

Filter used on EnVision Multilabel Reader:

- Absorbance: Photometric 595nm,
- Excitation: 485nm FITC,
- Emission: 535nm FITC,
- Mirror module: FITC (403) on top.

10µl of antibiotics was added to each medium.

Characterization of *yeaRp* dynamic range in LB

The concentrations used for the characterization of *yeaRp* was from 0 to 50 mM nitrate, with intervals of 10 mM.

Expected final nitrate concentration (mM)	**Actual final nitrate concentration (mM)**	**LB (ml)**	**1M KNO_3 (µl)**
0	0	10	0
10	9.89	10	100
20	19.58	10	200
30	29.10	10	300
40	38.42	10	400
50	47.57	10	500

The concentration of the characterization of *yeaRp* was from 0 to 10 mM of nitrate, with intervals of 2 mM.

Expected final nitrate concentration (mM)	**Actual final nitrate concentration (mM)**	**LB (ml)**	**1M KNO_3 (µl)**
0	0	10	0
2	1.99	10	20
4	3.98	10	40
6	5.96	10	60
8	7.93	10	80
10	9.89	10	100

Growth Medium: M9

Characterization of the promoter dynamic range in M9 minimal medium

The concentrations used for the characterization of *yeaRp* was from 0 to 2000 µM nitrate, with 10 folds increase for each interval.

Expected final nitrate concentration (µM)	**Actual final nitrate concentration (µM)**	**M9 (ml)**	**1M KNO_3 (µl)**
0	0	10	0
20	19.89	10	0.2
200	199.76	10	2
2000	1994.02	10	20

The concentrations of the characterization of *yeaRp* was from 0 to 500 µM of nitrate, with intervals of 100 µM.

Expected final nitrate concentration (µM)	**Actual final nitrate concentration (µM)**	**M9 (ml)**	**1M KNO_3 (µl)**
0	0	10	0
100	99.89	10	1
200	199.76	10	2
300	299.61	10	3
400	399.44	10	4
500	499.25	10	5

Reference:

Sambrook, J., & Russell, D. W. (2001). Molecular cloning. A laboratory manual. Third. *Cold pring Harbor Laboratory Press, New York*.